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14. ABSTRACT

The research involves laboratory studies utilizing xenograft models to test the hypothesis that targeting a member of the ETS transcription factor family with small molecules such as YK-4-279 may effectively treat prostate cancers. In year one Dr. Morrissey was to obtain internal Environmental Health and Safety (EH&S), Institutional Review Board (IRB), Institutional Animal Care and Use Committee (IACUC), Human Research Protections Office (HRPO) and Animal Care and Use Review Office (ACURO) approval. During this time Dr. Aykut Uren at Georgetown University was to screen more potential ETS transcription factor inhibitors for use in animal studies. Dr. Uren was to meet with and supply Dr. Morrissey with the inhibitor. All approvals have been applied for and obtained except for ACURO approval, which we have applied for and are currently awaiting. Dr. Uren has met with and delivered the inhibitor to Dr. Morrissey. The animal studies have been designed. Once ACURO approval is obtained the animal studies will begin.

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Table of Contents

Page

Introduction

A group of chromosomal translocations were recently discovered in prostate cancer that fuses the 5` region of *TMPRSS2* (a serine protease) gene to the 3` region of *ETS* transcription factor genes (1). *TMPRSS2* is an androgen responsive gene and contributes only its promoter region and usually a very short exon-1 (2, 3). This causes aberrant expression of an ETS transcription factor in response to androgen. The most common *ETS* member involved in prostate cancer chromosomal translocations is *ERG* but other members such as *ETV1, ETV4* and *ETV5* have been also observed (4, 5). The more aggressive prostate cancers often contain these translocations, thus potentially increasing their utility as both diagnostic and prognostic marker (6-8). Cell culture and transgenic animal models suggest that increased expression of ETS members, as a result of the chromosomal translocations, increase cell invasion without affecting the proliferative potential (9-11). However, in some xenograft models reducing expression of TMPRSS2-ERG protein slows down prostate cancer growth (12, 13). Therefore, ETS proteins emerge as potential novel targets for treatment of primary and/or metastatic disease in prostate cancer.

We developed small molecule inhibitors that target protein products of chromosomal translocations containing ETS transcription factors (14). We further established that our lead compound, YK-4-279, directly binds to both ERG and ETV1 proteins (15). YK-4-279 inhibits ERG and ETV1 mediated transcriptional activity and subsequent cellular invasive phenotype of prostate cancer cell lines. These effects were only observed in prostate cancer cell lines containing ETS chromosomal translocations such as VCaP and LNCaP and absent in the PC3 prostate cancer cell line that does not contain any ETS chromosomal translocations. Expression of ERG in PC3 cells from an expression vector sensitized them to YK-4-279 and inhibiting ERG expression in VCaP resulted in resistance to YK-4-279 effect (15). Therefore, we hypothesize that targeting ETS family of transcription factors by small molecules will inhibit malignant phenotypes of human prostate cancer cells.

Body

The research involves laboratory studies utilizing xenograft models to test the hypothesis that targeting a member of the ETS transcription factor family with small molecules such as YK-4-279 may effectively treat prostate cancers. In year 1 Dr. Uren was to screen ETS transcription factors to test in animal models of prostate cancer at the University of Washington. Dr. Morrissey was to obtain all approvals to start the animal studies in year 2, meet with Dr. Uren to discuss the fine details of the animal studies and obtain inhibitor from Dr. Uren for the animal studies.

Dr. Morrissey received a 2% effort in year 1.

Key Research Accomplishments

1. Dr. Uren has screened and tested a variety of ETS transcription factor inhibitors
2. Dr. Uren provided inhibitor to Dr. Morrissey
3. The animal studies have been designed
4. Dr. Morrissey has applied for all approvals and is awaiting ACURO approval

No research studies were designated to Dr. Morrissey in year 1.

Reportable Outcomes

In year one Dr. Morrissey was to obtain internal Environmental Health and Safety (EH&S), Institutional Review Board (IRB), Institutional Animal Care and Use Committee (IACUC), Human Research Protections Office (HRPO) and Animal Care and Use Review Office (ACURO) approval. During this time Dr. Aykut Uren at Georgetown University was to screen more potential ETS transcription factor inhibitors for use in animal studies. Dr. Uren was to meet with and supply Dr. Morrissey with the inhibitor. All approvals have been applied for and obtained except for ACURO approval, which we have applied for and are currently awaiting. Dr. Uren has met with Dr. Morrissey the details of the animal studies have been worked out and Dr. Uren has delivered the inhibitor to Dr. Morrissey.

Conclusion

Once ACURO approval is obtained the animal studies will begin at the University of Washington. We have no publications to report.

References

1. Tomlins SA, Rhodes DR, Perner S, Dhanasekaran SM, Mehra R, Sun XW, et al. Recurrent fusion of TMPRSS2 and ETS transcription factor genes in prostate cancer. Science. 2005;310:644-8.
2. Kumar-Sinha C, Tomlins SA, Chinnaiyan AM. Recurrent gene fusions in prostate cancer. Nat Rev Cancer. 2008;8:497-511.
3. Narod SA, Seth A, Nam R. Fusion in the ETS gene family and prostate cancer. Br J Cancer. 2008;99:847-51.
4. Helgeson BE, Tomlins SA, Shah N, Laxman B, Cao Q, Prensner JR, et al. Characterization of TMPRSS2:ETV5 and SLC45A3:ETV5 gene fusions in prostate cancer. Cancer Res. 2008;68:73-80.
5. Tomlins SA, Mehra R, Rhodes DR, Smith LR, Roulston D, Helgeson BE, et al. TMPRSS2:ETV4 gene fusions define a third molecular subtype of prostate cancer. Cancer Res. 2006;66:3396-400.
6. Demichelis F, Fall K, Perner S, Andren O, Schmidt F, Setlur SR, et al. TMPRSS2:ERG gene fusion associated with lethal prostate cancer in a watchful waiting cohort. Oncogene. 2007;26:4596-9.
7. Laxman B, Tomlins SA, Mehra R, Morris DS, Wang L, Helgeson BE, et al. Noninvasive detection of TMPRSS2:ERG fusion transcripts in the urine of men with prostate cancer. Neoplasia. 2006;8:885-8.
8. Wang J, Cai Y, Ren C, Ittmann M. Expression of variant TMPRSS2/ERG fusion messenger RNAs is associated with aggressive prostate cancer. Cancer Res. 2006;66:8347-51.
9. Carver BS, Tran J, Chen Z, Carracedo-Perez A, Alimonti A, Nardella C, et al. ETS rearrangements and prostate cancer initiation. Nature. 2009;457:E1; discussion E2-3.
10. Klezovitch O, Risk M, Coleman I, Lucas JM, Null M, True LD, et al. A causal role for ERG in neoplastic transformation of prostate epithelium. Proc Natl Acad Sci U S A. 2008;105:2105-10.
11. Tomlins SA, Laxman B, Dhanasekaran SM, Helgeson BE, Cao X, Morris DS, et al. Distinct classes of chromosomal rearrangements create oncogenic ETS gene fusions in prostate cancer. Nature. 2007;448:595-9.
12. Sun C, Dobi A, Mohamed A, Li H, Thangapazham RL, Furusato B, et al. TMPRSS2-ERG fusion, a common genomic alteration in prostate cancer activates C-MYC and abrogates prostate epithelial differentiation. Oncogene. 2008;27:5348-53.
13. Wang J, Cai Y, Yu W, Ren C, Spencer DM, Ittmann M. Pleiotropic biological activities of alternatively spliced TMPRSS2/ERG fusion gene transcripts. Cancer Res. 2008;68:8516-24.
14. Erkizan HV, Kong Y, Merchant M, Schlottmann S, Barber-Rotenberg JS, Yuan L, et al. A small molecule blocking oncogenic protein EWS-FLI1 interaction with RNA helicase A inhibits growth of Ewing's sarcoma. Nature medicine. 2009;15:750-6.
15. Rahim S, Beauchamp EM, Kong Y, Brown ML, Toretsky JA, Uren A. YK-4-279 Inhibits ERG and ETV1 Mediated Prostate Cancer Cell Invasion. PloS one. 2011;6:e19343.